

WHAT IS CLAIMED IS:

1                   1.       A method of oxidizing a phosphite ester linkage in a nucleic acid  
2 array to a phosphate linkage, comprising contacting said phosphite ester linkage with a  
3 solution of from about 0.005 M to about 0.05 M iodine in a mixture of water and organic  
4 solvent.

1                   2.       A method of preparing a nucleic acid array on a support, wherein  
2 each nucleic acid occupies a separate known region of the support, said synthesizing  
3 comprising:

4                   (a) activating a region of the support;

5                   (b) attaching a nucleotide to a first region, said nucleotide having a  
6 masked reactive site linked to a protecting group;

7                   (c) repeating steps (a) and (b) on other regions of said support whereby  
8 each of said other regions has bound thereto another nucleotide comprising a masked  
9 reactive site link to a protecting group, wherein said another nucleotide may be the same  
10 or different from that used in step (b);

11                  (d) removing the protecting group from one of the nucleotides bound to  
12 one of the regions of the support to provide a region bearing a nucleotide having an  
13 unmasked reactive site;

14                  (e) binding an additional nucleotide to the nucleotide with an unmasked  
15 reactive site;

16                  (f) repeating steps (d) and (e) on regions of the support until a desired  
17 plurality of nucleic acids is synthesized, each nucleic acid occupying separate known  
18 regions of the support;

19                  wherein said attaching and said binding are each made by covalently forming a  
20 phosphite triester linkage between said nucleotides and said unmasked reactive site and  
21 further comprising oxidizing said phosphite triester linkage to a phosphate triester linkage  
22 with a solution of from about 0.005 M to about 0.05 M iodine in an aqueous solvent  
23 mixture.

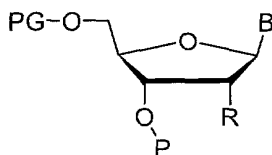
1                   3.       A method in accordance with claim 2, wherein said synthesizing  
2 comprises the sequential steps of:

3                   a) removing a photoremoveable protecting group from at least a first area  
4 of a surface of a substrate, said surface comprising immobilized nucleotides on said

5 surface, said nucleotides capped with a photoremovable protective group, without  
 6 removing a photoremoveable protecting group from at least a second area of said surface;  
 7 b) simultaneously contacting said first area and said second area of said  
 8 surface with a first nucleotide to couple said first nucleotide to said immobilized  
 9 nucleotides in said first area, and not in said second area, said first nucleotide capped with  
 10 said photoremovable protective group;  
 11 c) removing a photoremoveable protecting group from at least a part of  
 12 said first area of said surface and at least a part of said second area;  
 13 d) simultaneously contacting said first area and said second area of said  
 14 surface with a second nucleotide to couple said second nucleotide to said immobilized  
 15 nucleotides in at least a part of said first area and at least a part of said second area;  
 16 e) performing additional irradiating and nucleotide contacting and  
 17 coupling steps so that a matrix array of at least 100 nucleic acids having different  
 18 sequences is formed on said support;  
 19 with the proviso that the coupling steps further comprise oxidizing an  
 20 initially formed phosphite ester linkage to a phosphate ester linkage using from about  
 21 0.005 M to about 0.05 M iodine in an aqueous solvent mixture.

1 4. A method in accordance with claim 3, wherein said aqueous  
 2 solvent mixture comprises iodine in an amount of about 0.02 M.

1 5. A method in accordance with claim 3, wherein said nucleotides  
 2 have the formula:



3 wherein

4 B is a member selected from the group consisting of natural or unnatural  
 5 adenine, natural or unnatural guanine, natural or unnatural thymine,  
 6 natural or unnatural cytosine, and natural or unnatural uracil;  
 7 R is a member selected from the group consisting of hydrogen, hydroxy,  
 8 protected hydroxy, halogen and alkoxy;  
 9 P is a phosphoramidite group; and  
 10 PG is a photoremoveable protected group. .

1                   6.       A method in accordance with claim 5, wherein B is selected from  
2       the group consisting of adenine, guanine, cytosine and thymine and R is hydrogen.

1                   7.       A method in accordance with claim 5, wherein said array  
2       comprises at least 10 different nucleic acids.

1                   8.       A method in accordance with claim 5, wherein said array  
2       comprises at least 100 different nucleic acids.

1                   9.       A method in accordance with claim 5, wherein said array  
2       comprises at least 1000 different nucleic acids.

1                   10.      A method in accordance with claim 5, wherein said array  
2       comprises at least 10,000 different nucleic acids.

1                   11.      A method in accordance with claim 5, wherein said array  
2       comprises at least 100,000 different nucleic acids.

1                   12.      A method in accordance with claim 5, wherein each different  
2       nucleic acid is in a region having an area of less than about 1 cm<sup>2</sup>.

1                   13.      A method in accordance with claim 5, wherein each different  
2       nucleic acid is in a region having an area of less than about 1 mm<sup>2</sup>.

1                   14.      A method in accordance with claim 5, wherein said solution is  
2       about 0.02 M iodine in a mixture of water, pyridine and THF.

1                   15.      A method in accordance with claim 5, wherein B is selected from  
2       the group consisting of adenine, guanine, cytosine and thymine, R is hydrogen, and said  
3       solution is about 0.02 M iodine in a mixture of water, pyridine and THF.

1                   16.      A method in accordance with claim 5, wherein B is selected from  
2       the group consisting of adenine, guanine, cytosine and thymine, R is hydrogen, PG is  
3       MeNPOC and said solution is about 0.02 M iodine in a mixture of water, pyridine and  
4       THF.

1                   17.      A method in accordance with claim 5, wherein B is selected from  
2       the group consisting of adenine, guanine, cytosine and thymine, R is hydrogen, PG is

- 3 MeNPOC, P is  $-P(OCH_2CH_2CN)N(iPr)_2$  and said solution is about 0.02 M iodine in a
- 4 mixture of water, pyridine and THF.

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